determining whether said test nucleic acid sample contains a sequence variance.

REMARKS

The present claims recite methods of using a probe derived from a sex chromosome or a somatic cell hybrid to determine whether a nucleic acid sample contains a sequence variance. The present claims include amended versions of claims 22, 23 (which is now presented as claim 51), 64, and 65 (which are now presented as claims 49 and 50) from parent application U.S.S.N. 09/697,097 as well as new claims 46-48, and are believed to be in condition for allowance. Specifically, in the Office Action received in connection with the parent application (mailed August 2, 2001; "Office Action"), rejections were raised regarding novelty and obviousness. Each of these rejections is addressed below in the order in which they appeared in the Office Action.

Support for the Amendments

Applicant has amended claim 22 and added new claims 46-51. Claim 22 has been amended to clarify that the probes and detection techniques are utilized for the purpose of analyzing a test sample to determine whether that sample includes a sequence variance. This amendment finds support throughout the specification, for example, at page 1, lines 27-28, and page 11, lines 10-11. These amendments serve to place the invention in its intended context, relating to the discovery and detection of new variances and discovery of their organization, as described in Applicant's specification at page 1, lines 10-14.

New claims 46-48 specify that the probe is derived from a cell having only one parental copy of an X chromosome and/or only one parental copy of a Y chromosome or that the probe is derived from a hemizygous cell (as disclosed, for example, at page 6, lines 4-20, page 22, lines 14-17, and page 23, lines 2-6, of the specification).

And new claims 49 and 50 (which correspond to claims 64 and 65 of parent application U.S.S.N. 09/697,097) find support throughout the application, for example, at

page 23, lines 19-24, and at page 32, lines 16-19, where the use of human sex chromosomes or human test nucleic acids is described.

New claim 51 (which corresponds to claim 23 of the parent application) recites a method for detecting a sequence variance using a probe derived from a somatic cell hybrid (as disclosed, for example, on pages 27-29 of the specification).

These amendments add no new matter.

Rejections from Office Action in Parent Application

Each of the rejections in the Office Action received in the parent application is addressed below as they pertain to present claims 22 and 46-51.

Prior claim 22 was rejected, under 35 U.S.C. § 102(a), as being anticipated by Halverson *et al.* (U.S. Patent No. 5,707,809). As applied to the corresponding present claims, this rejection is respectfully traversed. Specifically, claims 22 and 46-50 are now directed to methods for determining whether a test nucleic acid sample contains a sequence variance by determining whether there is a nucleotide mismatch between the test nucleic acid and a probe derived from a sex chromosome. Nowhere in Halverson is such a technique for detecting a sequence variance disclosed.

The focus of Halverson is determining the gender of a bird prior to the development of external sexual characteristics. In particular, Halverson discloses methods for determining whether an avian DNA sample contains two Z chromosomes and thus is from a male bird, or contains Z and W chromosomes and thus is from a female bird. These methods involve hybridizing a probe to one or both avian sex chromosomes. Because the focus of Halverson is determining what sex chromosomes are present in the sample rather than determining what mutations are present in the sex chromosomes, Halverson does not teach or suggest using the probe as a standard to determine whether a sex chromosome has a sequence variation or mutation relative to the

probe. Thus, none of the Halverson methods involve detecting a nucleotide mismatch due to a lack of complementarity between a sex chromosome and the probe, as required by the present claims.

In one approach described by Halverson, a probe that hybridizes exclusively to one of the two avian sex chromosomes is used to determine the gender of the bird based on the hybridization intensity of the probe to the DNA sample. For example, if a probe specific for the W chromosome hybridizes to the DNA sample, then the DNA sample is from a female bird (genotype WZ). Alternatively, if the probe does not hybridize to the DNA sample, then the DNA sample is from a male bird (genotype ZZ). In another embodiment of this method, a probe specific for the Z chromosome binds with approximately double the hybridization intensity to a DNA sample from a male bird (due to the presence of two Z chromosomes) than to a sample from a female bird. As noted above, Halverson does not teach or suggest the detection of a nucleotide mismatch between the probe and the DNA sample. For example, the hybridization of the probe to a sex chromosome may occur if there are 0, 1, 2, or more nucleotide mismatches between the probe and the sex chromosome. As indicated in column 5, lines 23 and 24, of Halverson, "[p]robes may have either complete or partial sequence identity to the sex specific sequence." Thus, measuring the hybridization intensity as taught by Halverson does not determine whether or not any nucleotide mismatches are present, as required by the present claims.

In the alternative approach, a probe that hybridizes to both sex chromosomes is used for hybridization to a restriction enzyme-cleaved DNA sample. Differences in the polynucleotide sequence between the two avian chromosomes result in one or more differences in the location or number of restriction enzyme cleavage sites between the two avian chromosomes. As illustrated in Table 1 of Halverson, these differences in restriction enzyme cleavage sites result in differences between the size of the cleavage products of the restriction enzyme-digested Z chromosome and the size of the cleavage

products of the restriction enzyme-digested W chromosome. Thus, the sizes of cleavage products that hybridize to the probe are indicative of which avian chromosomes are present in the DNA sample. As one skilled in the art would appreciate, this method is based on sequence differences between the two avian sex chromosomes, not nucleotide mismatches between the probe and a sex chromosome. Nowhere does Halverson teach or suggest detecting a nucleotide mismatch between the probe and a test nucleic acid (*e.g.*, a sex chromosome), as required by the present claims. As noted above, the hybridization of a cleavage product to the probe does not indicate whether or not any nucleotide mismatches are present between the cleavage product and the probe.

As Halverson fails to disclose the critical element of determining whether there is a nucleotide mismatch between the sex chromosome and the probe, this rejection as applied to the present claims should be withdrawn.

In addition, Applicant further notes that Halverson does not teach the use of probes derived from human sex chromosomes, cells having only one parental copy of an X or Y chromosome, hemizygous cells, or human nucleic acid test samples, as required by new claims 46-50. These claims are novel over the Halverson reference for these reasons as well.

Regarding the other novelty rejection, prior claim 23 was rejected, under 35 U.S.C. § 102(e), as being anticipated by Whiteley *et al.* (U.S. Patent No. 5,962,223). As applied to the corresponding present claim 51, this rejection is respectfully traversed.

Claim 51 recites a sequence variance detection method that uses a probe derived from a somatic cell hybrid. The Office asserts that the limitation that the somatic cell hybrid is formed from the fusion of a cell or chromosome to a recipient cell is not recited in prior claim 23. In the interest of expediting prosecution, Applicant has included this limitation in present claim 51. For the record, Applicant notes that it is not necessary to

add the definition of a somatic cell hybrid to the claim to clearly recite the present method because one skilled in art would know that a somatic cell hybrid is formed from the fusion of a cell or chromosome to a recipient cell (see, for example, page 27, lines 1-7, of the specification). Applicant further notes that the inclusion of the limitation defining a somatic cell hybrid in no way alters the scope of the present claim.

Nowhere is the claimed method disclosed by the Whiteley reference. In contrast to the Office's assertion that Example 2 discloses the generation of a probe from a somatic cell hybrid, Applicant notes that the probes described in Example 2 were synthesized using a DNA synthesizer (column 8, lines 41-57, and column 11, lines 20-22). Whiteley does not teach or suggest the possibility of generating a probe from a somatic cell hybrid.

Moreover, the methods described by Whiteley involve the hybridization of both a diagnostic probe and a contiguous probe to a test nucleic acid. For the detection of a mutation of interest in a test nucleic acid, Whiteley teaches use of a probe with a sequence complementary to the region containing the mutation to determine whether the probe hybridizes to the test nucleic acid, thereby determining whether the test nucleic acid contains the mutation of interest (column 2, lines 43-49). Whiteley does not disclose the detection of a nucleotide mismatch between the probe and a test nucleic acid due to the presence of a nucleotide in the probe that is not complementary to the corresponding nucleotide in the test nucleic acid, as required by the present claims.

As Whiteley fails to disclose multiple important elements of Applicant's claim 51, the rejection of this claim should be withdrawn.

With respect to the obviousness rejection, prior claims 22, 64, and 65 were rejected, under 35 U.S.C. § 103(a), as being unpatentable over Halverson in view of Ward *et al.* (U.S. Patent No. 6,007,994). This rejection of the corresponding present claims 22 and 46-50 is also respectfully traversed. As noted above, Halverson does not

disclose or suggest the use of a probe derived from a sex chromosome to detect a sequence variance. The methods disclosed by Ward involve the hybridization of a probe to a test sample to determine which chromosomes are present in the sample. Ward does not disclose the detection of a nucleotide mismatch between the probe and the test sample, as required by the present claims. As neither Halverson nor Ward, alone or in combination, teach or suggest the methods recited in the present claims, this obviousness rejection should be withdrawn.

Conclusion

Applicant submits that this case is now in condition for allowance, and such action is respectfully requested. A marked-up version indicating the amendments made to the claims, as required by 37 C.F.R. § 1.121(c)(1)(ii), is enclosed.

If there are any charges, or any credits, please apply them to Deposit Account No. 03-2095.

Respectfully submitted,

Date: 7 January 2002

Karen L. Elbing, Ph.D.
Reg. No. 35,238

Clark & Elbing LLP
176 Federal Street
Boston, MA 02110
Telephone: 617-428-0200
Facsimile: 617-428-7045

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	Vincent P. Stanton, Jr.	Art Unit:	Not yet assigned
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Title:	PROBES FOR VARIANCE DETECTION		

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Washington, D.C. 20231

Version with Markings to Show Changes Made

A marked-up version of claim 22 and new claims 46-51 are presented below.

22. (Amended) A method for analyzing a test nucleic acid sample to determine whether it contains a sequence variance [detecting a nucleotide mismatch in a nucleic acid sample], said method comprising the steps of:

(a) obtaining a nucleic acid probe that is complementary to a sex chromosome or segment thereof; [providing a nucleic acid probe derived from a sex chromosome;]

(b) forming a duplex between said test nucleic acid sample and said probe; and

(c) analyzing whether [determining if] said duplex contains a nucleotide mismatch, thereby determining whether said test nucleic acid sample contains a sequence variance.

46. (New) The method of claim 22, wherein said probe is derived from a cell having only one parental copy of an X chromosome or only one parental copy of a Y chromosome.

47. (New) The method of claim 22, wherein said probe is derived from a cell having only one parental copy of an X chromosome and only one parental copy of a Y chromosome.

48. (New) The method of claim 22, wherein said probe is derived from a hemizygous cell.

49. (New) The method of claim 22, wherein said sex chromosome is from a human.

50. (New) The method of claim 22, wherein said test nucleic acid sample is from a human.

51. (New) A method for analyzing a test nucleic acid sample to determine whether it contains a sequence variance, said method comprising the steps of:

(a) obtaining a nucleic acid probe from a somatic cell hybrid, said probe being complementary to a chromosome or segment thereof, wherein only one allele of said chromosome or segment thereof is present in said somatic cell hybrid, and wherein said somatic cell hybrid is formed from the fusion of a cell or chromosome to a recipient cell;

(b) forming a duplex between said test nucleic acid sample and said probe; and

(c) analyzing whether said duplex contains a nucleotide mismatch, thereby determining whether said test nucleic acid sample contains a sequence variance.